American Journal of Medical Genetics 68:270–278 (1997)

Cardiovascular Malformations in Smith-Lemli-Opitz Syndrome

Angela E. Lin,[1*] Holly H. Ardinger,[2] Robert H. Ardinger, Jr.,[3] Christopher Cunniff,[4] and Richard I. Kelley[5]

[1] *Westwood, Massachusetts*
[2] *Division of Genetics, Department of Pediatrics University of Kansas, Kansas City*
[3] *Division of Cardiology, University of Kansas, Kansas City*
[4] *Section of Medical and Molecular Genetics, Department of Pediatrics, University of Arizona, Tuscon*
[5] *The Kennedy Krieger Institute, Department of Pediatrics and Johns Hopkins University, Baltimore, Maryland*

We reviewed 215 patients (59 new, 156 from the literature) with Smith-Lemli-Opitz syndrome (SLOS), and found that 95 (44%) had a cardiovascular malformation (CVM). Classifying CVMs by disordered embryonic mechanisms, there were 5 (5.3%) class I (ectomesenchymal tissue migration abnormalities), 56 (58.9%) class II (abnormal intracardiac blood flow), 25 (26.3%) class IV (abnormal extracellular matrix), and 5 (5.3%) class V (abnormal targeted growth). Comparing the frequencies of *individual* CVMs in this series with a control group (the Baltimore-Washington Infant Study), there were 6 individual CVMs which showed a significant difference from expected values. When frequencies of CVMs in SLOS were analyzed by mechanistic *class*, classes IV and V were significantly more frequent, and class I significantly less frequent, than the control group. Although CVMs in SLOS display mechanistic heterogeneity, with an overall predominance of class II CVMs, the developmental error appears to favor alteration of the cardiovascular developmental mechanisms underlying atrioventricular canal and anomalous pulmonary venous return. This information should assist the clinical geneticist evaluating a patient with possible SLOS, and should suggest research direction for the mechanisms responsible for the SLOS phenotype. Am. J. Med. Genet. 68:270–278, 1997. © 1997 Wiley-Liss, Inc.

KEY WORDS: MCA/MR syndrome; cardiovascular malformation; congenital heart defect; mechanistic classification; atrioventricular canal; anomalous pulmonary venous return; Smith-Lemli-Opitz syndrome

INTRODUCTION

Smith-Lemli-Opitz syndrome (SLOS) is a well-known multiple congenital anomaly/mental retardation syndrome comprising a distinct facial appearance (microcephaly, bitemporal narrowing, ptosis, anteverted nares, and apparently low-set ears), cleft palate, syndactyly of the second and third toes, hypoplastic external genitalia, and multiple internal abnormalities involving the skeleton, eyes, lungs, kidney, brain, and gastrointestinal system. In addition to the usual moderate-to-severe mental retardation, there is often substantial hypotonia, failure to thrive, and reduced life span.

Diagnostic criteria have been proposed for the more severely affected type II patients (i.e., presence of at least three of the following: cleft palate, polydactyly, cardiovascular malformation (CVM), cataract, small tongue, severe genital ambiguity, characteristic clinical course; plus, one or more of the less common or less severe abnormalities: Hirschsprung disease, renal abnormalities, large adrenals, pancreatic islet cell hypertrophy, unilobate lungs, 2–3 toe syndactyly, redundant neck skin, short limbs, facial hemangioma, joint contractures) [Curry et al., 1987]. Whether or not SLOS types I and II represent distinct clinical entities has been questioned.

For the "typical" SLOS patient, minimal phenotypic diagnostic criteria remain problematic, and a formal definition is lacking to diagnose confidently milder forms of SLOS or those overlapping with other syndromes on a clinical basis [Verloes, 1995].

*Correspondence to: Angela E. Lin, M.D., 272 Dover Road, Westwood, MA 02090-2414.

Received 31 May 1996; Accepted 2 July 1996

However, with the recent discovery of the same abnormality of sterol metabolism in all forms of SLOS [Tint et al., 1994; Irons et al., 1994], it is now possible to "redefine" SLOS biochemically. One can diagnose definitively some patients in whom the clinical diagnosis is unclear.

The first reported CVM in SLOS was described as "compatible with tetralogy of Fallot" in the sixteenth patient [Park et al., 1968]. In two early series of 40 [Robinson et al., 1971] and 63 [Johnson, 1975] patients with SLOS, CVMs were noted in approximately 20% without a predominant type [Lowry, 1983]. Cardiovascular malformations were more common (80%) in the more severe form, SLOS type II [Curry et al., 1987; Verloes, 1995].

To determine if a particular type of embryonic class of CVM was characteristic of SLOS, we reviewed the frequency and pattern of CVMs in SLOS in published cases and in new cases diagnosed by biochemical criteria. Such information could assist in the recognition of patients to be biochemically screened for this disorder, and in directing research toward how biochemical abnormality produces the multiple phenotypic abnormalities seen in SLOS.

PATIENTS AND METHODS

We reviewed previously reported patients from an extensive bibliography [Opitz et al., 1994], which was updated to 1995 to include 156 patients (Table I). We also include data on 59 new patients, 58 of whom were referred for biochemical analysis to one of the authors (R.K.) at the Kennedy Krieger Institute [Cunniff et al., 1997].

Excluded were literature patients annotated as "not verified" [Opitz et al., 1994], inadequately described in brief reports or abstracts, or lacking sufficient traits based on our own review. We excluded patients in whom there was extensive overlap with other syndromes, such as Meckel syndrome [Thompson and Baraitser, 1987; Casamassima et al., 1987]; Pallister-Hall syndrome [Donnai et al., 1987]; Gardner-Silengo-Wachtel syndrome [Greenberg et al., 1987]; and holoprosencephaly-polydactyly (pseudotrisomy 13) syndrome [Verloes et al., 1991]. We also excluded patients with atypical SLOS who had a chromosome abnormality, but lacked biochemical confirmation, such as "Q plus" with Seckel syndrome anomalies [Ballesta et al., 1974], de novo t(1p22;2q23) [Chitayat et al., 1989], direct duplication 2p14→2p23 [Yunis et al., 1979], de novo 2q+ [Donnenfeld et al., 1987], deletion 4p [Hill et al., 1991], t(7;17)(q34;p13.1) with SLOS and Miller-Dieker syndrome anomalies [Berry et al., 1989], and del Xp11 [patient 2, McKeever and Young, 1990].

Included in our review were 2 patients with rearrangements involving a putative SLOS locus on 7q32, i.e., mat t(6p21;7q32) [patient 7, Curry et al., 1987] and t(7q32;20q13) [Wallace et al., 1994], the latter having been shown to have the characteristic sterol abnormality.

Biochemical confirmation of the diagnosis of SLOS was defined as elevated levels of 7-dehydrocholesterol from any tissue as performed at one of two laboratories, as described previously [Tint et al., 1994; Kelley, 1995].

We used the guidelines of Verloes [1995] to extract data from reports. Thus, easily observed manifestations (e.g., cleft palate, polydactyly) not stated in text were listed as absent (−). We listed as "unknown" minor anomalies (e.g., epicanthus) not stated in the text, and internal visceral anomalies (e.g., central nervous system malformations), unless autopsy or specific diagnostic tests were done. Pyloric stenosis and Hirschsprung disease were recorded as absent in pa-

TABLE I. Summary of Clinical Manifestations in 215 Patients (156 From the Literature, 59 New) With SLOS*

	Literature patients			New patients		
Manifestation	No. informative	No. present	(%)	No. informative	No. present	(%)
Phenotypic gender	137			59		
Female		69	(50)		32	(54)
Male		65	(48)		21	(36)
Ambiguous		3	(2)		6	(10)
Discrepant phenotype/karyotype female/46,XY	113	28	(25)	59	11	(19)
Face						
Cataract	149	34	(23)	58	13	(22)
Cleft palate/uvula	144	66	(46)	58	27	(47)
Limbs						
2/3 Toe syndactyly	149	121	(81)	58	58	(100)
Polydactyly	149	67	(45)	59	31	(52)
Gastrointestinal						
Hirschsprung disease	147	18	(12)	53	8	(15)
Pyloric stenosis	131	14	(11)	53	4	(7)
Genital anomaly	136	93	(68)	59	29	(49)
Kidney anomaly	101	50	(49)	25	10	(40)
Lungs, unilobate	79	25	(32)	9	3	(33)
CNS anomaly	82	46	(56)	18	3	(16)
Psychomotor retardation	93	93	(100)	41	41	(100)
Mild		3	(3)		18	(44)
Moderate-to-severe		90	(97)		23	(56)

* References 1, 2, 4, 6–8, 10–14, 18, 20–22, 24, 27–34, 37–40, 42–49, 51–59, 61–71, 73–80, 82, 83, 87, 88, 90–93, 95, 96, 99, 101, 102. CNS, central nervous system.

tients who were at least age 6 months and who had no gastrointestinal symptoms other than nonspecific growth failure.

A CVM was defined as a congenital structural heart defect. Excluded were patients with cardiomyopathy, arrhythmia, patent ductus arteriosus (PDA) in premature infants <36 weeks of gestation or <4 days of life, and nonspecific murmurs. In 3 patients a CVM was reported as present, but was unsubstantiated in the text [patients 6 and 7, Dallaire and Fraser, 1969; patient 19, Curry et al., 1987].

We categorized CVMs by specific anatomic defect, using a classification based on postulated embryonic mechanisms [Clark, 1990]. A ventricular septal defect (VSD) whose anatomic location was not described was classified as "VSD, type not specified." Because another prevalence study [Ferencz et al., 1987] did not specify VSD location, we included those VSDs not otherwise specified in the calculation of class II VSD (membranous). Similarly, if the location of an atrial septal defect was not described, it was assumed to be a secundum type.

Apart from patients who died in infancy, all patients were considered to have mental retardation, except the patients of Lowry and Yong [1980], and of Joseph et al. [1987], who all had borderline abnormal intelligence.

RESULTS

A summary of phenotypic manifestations of SLOS patients (215, total; 156 from the literature, 59 new) is presented in Table I. Figure 1 shows the typical appearance of the first new patient (patient ID #157).

Fig. 1. Patient ID #157 (new patient 1). Full-length photograph shows typical facial appearance, multiple contractures, and postaxial polydactyly of hands and feet.

In addition to the phenotypic data presented in Table I, there were 72 patients (14 from the literature, 58 new) who had biochemical testing supporting the clinical diagnosis of SLOS, i.e., increased 7-dehydrocholesterol in plasma, tissues, or cultured cells.

Of 215 patients, a CVM was reported in 95 (44%), and was sufficiently well-defined in 91 patients to be classified. There were 74 (47%) literature and 21 (36%) new patients with a CVM.

The specific CVMs identified in this review are listed in Table II. Table III compares the distribution of CVM classes in this study with the Baltimore-Washington Infant Study (BWIS), a large population-based study on CVM prevalence [Ferencz et al., 1987]. The frequency of class II CVMs was similar, i.e., 58.9% in this study compared with 64% in the BWIS. However, class IV CVMs (complete atrioventricular canal, primum type atrial septal defect, and common atrium) were more than twice as common, and class V CVMs (total and partial anomalous pulonary venous return) were almost three times as common in SLOS, compared with the BWIS. Class I CVMs (conotruncal and branchial arch defects) were approximately one fourth as common. Despite several patients with spleen anomalies, there were no cases of heterotaxy with complex CVMs.

When the frequencies of individual CVMs in SLOS were analyzed by a chi-square test, four types of CVM were significantly more frequent: secundum type atrial septal defect, patent ductus arteriosus, atrioventricular canal, and anomalous pulmonary venous return (Table IV). Two CVMs, VSD and tetralogy of Fallot, were significantly less frequent. The validity of the VSD analysis is based on the assumption that the VSDs of unspecified anatomic location were probably membranous (class II), and that VSDs listed as membranous were in that location.

When the CVMs were analyzed by groups, classes I, IV, and V maintained statistical significance (Table V). Class I CVMs were significantly less frequent in the SLOS group than in the BWIS group.

Of the 72 patients who had biochemical confirmation of the clinical diagnosis of SLOS, 26 (36.1%) had a CVM. The frequency of CVM classes in this group of biochemically confirmed patients was similar to what was observed in all patients (Table III), with the exception of class V CVMs, none of which was confirmed biochemically.

DISCUSSION

Cardiovascular malformations are common in SLOS and display mechanistic heterogeneity, with a predominance of class II CVMs. The increased proportion of class IV and V CVMs and decreased frequency of class I CVMs are intriguing.

Class IV CVMs are postulated to be due to an abnormality of myocardial mesenchymal cells and their ex-

TABLE II. Types of CVMs in SLOS Based on an Embryonic Mechanistic Classification (n = 95)*

Class	No. of patients	
I. Ectomesenchymal tissue migration abnormalities		5
A. Conotruncal septation defects		
TOF	2	
IAA, type B	1	
B. Abnormal conotruncal cushion position		
C. Branchial arch defects: aberrant subclavian artery, isolated	2	
II. Abnormal intracardiac blood flow		56
A. Ventricular septal defect, membranous		
Isolated	7	
With PAPVR	1	
With bicuspid pulmonic valve, PDA	1	
B. Left-sided cardiac anomalies		
Aortic stenosis	2	
Coarctation	3	
Hypoplastic left heart syndrome	3	
C. Right-sided anomalies		
ASD, secundum type	19	
Pulmonic stenosis	2	
Tricuspid atresia	1	
D. Patent ductus arteriosus		
Isolated	15	
With ASCA	1	
With RAA	1	
III. Cell death		0
IV. Extracellular matrix		25
A. Atrioventricular canal defects		
Complete AVC (2 had T/PAPVR in addition to AVC)	21	
ASD, primum type (1 had PAPVR in addition to ASD)	3	
Common atrium	1	
V. Abnormal targeted growth		5
A. Anomalous pulmonary venous return		
Total APVR (TAPVR with other CVM classified elsewhere, 1)	5	
Partial APVR (PAPVR with other CVM classified elsewhere, 3)	0	
VI. Situs and looping defects		0
Heterotaxy (4 spleen anomalies, 3 with CVM, none with heterotaxy)		
VII. Miscellaneous defects		0
Miscellaneous unclear cases (CVM reported, but data insufficient to classify)		4

* ASD, atrial septal defect; T/PAPVR, total/partial anomalous pulmonary venous return; AVC, atrioventricular canal; IAA, interrupted aortic arch; RAA, right aortic arch; TOF, tetralogy of Fallot.
List does not include every possible type of CVM, but only those which were reported as present.

TABLE III. Classes of CVMs in SLOS in This Study (Total and Those Confirmed Biochemically) Compared With Baltimore-Washington Infant Study (BWIS)*

	SLOS (present study)		BWIS, Ferencz et al., 1987 (%)
	Total n (%)	Biochemical confirmation n (%)	
CVM, reported	95 (44)	26 (36.1)	1,467 (excluding 27 cardiomyopathy)
Abnormality of mechanistic class			
I. Ectomesenchymal tissue migration	5.3	7.7	17
II. Intracardiac flow	58.9	69.2	64
III. Cell death	0	0	1
IV. Extracellular matrix	26.3	19.2	11
V. Targeted growth	5.3	0	2
VI. Situs and looping	0	0	3
VII. Miscellaneous, other	0	0	0
	4.2	3.8	2

*Modified from Clark [1990].

TABLE IV. Individual CVMs in SLOS Compared With Baltimore-Washington Infant Study†

	SLOS (95 CVMs)	BWIS (1,467 CVMs)	χ^2	P (1 df)
Class I TOF	2	138	5.38	<.025*
Class II				
VSD memb	9	393	14.52	<.001*
ASD 2°	19	112	20.60	<.001*
AoS	2	49	0.45	>.05
COA	3	101	2.06	>.05
HLHS	3	85	1.21	>.05
PS	2	105	3.65	>.05
PDA	17	39	85.22	<.001*
TAt	1	20	0.07	>.05
Class IV				
AVC	25	8.5	40.23	<.001*
Class V				
T/PAPVR	5	1.6	14.96	<.001*

† AoS, aortic stenosis; ASD, atrial septal defect; APVR, anomalous pulmonary venous return; AVC, atrioventricular canal; COA, coarctation; HLHS, hypoplastic left heart syndrome; PDA, patent ductus arteriosus; PS, pulmonic stenosis; TAt, tricuspid atresia; VSD memb, membranous ventricular septal defect.
* Statistically significant.

tracellular matrix that contributes to the endocardial cushions of the atrioventricular orifice and ventricular outflow tract [Clark, 1990]. Maldevelopment of the cushions results in the spectrum of defects ranging in severity from primum type atrial septal defect with cleft mitral valve, common atrium, inlet "canal-type" VSD, and transitional atrioventricular canal, to the complete common atrioventricular canal. In addition to these septal defects, some types of congenital nodular valvar dysplasia may be considered later-onset abnormalities of endocardial cushion organization.

Another common SLOS anomaly, Hirschsprung disease, may also be the result of a developmental error involving the extracellular matrix. Although Hirschsprung disease was initially attributed to a failure of neural crest cell migration to the gastrointestinal tract, recent evidence supports the hypothesis of appropriate migration to the gut but failure to penetrate bowel wall due to abnormal extracellular matrix in the affected bowel segment [Rudolph and Benaroch, 1995]. Based on this theory, one can hypothesize that the cholesterol abnormality in SLOS may alter the extracellular matrix, resulting in at least some of the anomalies found in SLOS patients. Since cholesterol usually does not play a great role in the extracellular matrix itself, cellular membranes and cell-to-cell interactions may be preferentially affected.

Pulmonary veins develop independently as outpouchings of the endothelial lined mesenchymal tissue in the lung buds, which coalesce to form the common pulmonary vein. Through an incompletely-understood process of targeted cell growth involving a signal from the left atrium, the veins fuse to the left atrium. Perhaps in a situation analogous to Hirschsprung disease, abnormal extracellular matrix in SLOS may alter this signal. Absorption of the common pulmonary vein results in four individual pulmonary veins. Class V CVMs have been attributed to absent or partial incorporation of the pulmonary veins into the left atrium, resulting in total or partial anomalous pulmonary venous return and cor triatriatum, respectively. The abnormal lung lobation common in SLOS also may interfere with the process of pulmonary vein targeted growth and incorporation.

The decreased frequency of class I (conotruncal and branchial arch) CVMs suggests that abnormal cholesterol synthesis in SLOS does not have a major effect on branchial arch development. This is supported by the lack of major craniofacial anomalies generally associated with branchial arch abnormalities, e.g., ear tags or structural ear anomalies, microtia, and pigmentary abnormality.

Because of the phenotypic overlap between SLOS and several other syndromes, the clinical geneticist

TABLE V. CVMs Grouped by Embryonic Class in SLOS Compared With Baltimore-Washington Infant Study (BWIS)

CVM class	SLOS (95 CVMs)	BWIS (1,467 CVMs)	χ^2	P(1 df)
I	5	245	8.93	<.005*
II	56	932	0.86	>.05
III	0	16	1.05	>.05
IV	25	167	21.00	<.001*
V	5	26	6.65	<.01*

* Statistically significant.

TABLE VI. CVMs in SLOS Compared With SLOS-Like Syndromes*

	Reference	Total	CVM present	CVM class I	II	IV	V	Miscellaneous
SLOS	Present study	215	44%	5%	59%	26%	5%	4%
Joubert	Saraiva and Baraitser, 1992	>100	Rare					
Oro-facial-digital (Varadi)	Gorlin et al., 1990	15	Rare		AoS	AVC		
Meckel	Salonen, 1984 Shen-Schwartz and Dave, 1988	>200	20%		90% ASD, PDA, BAV, HLHS			Polysplenia: complex CVM, VSD, NS
Pallister-Hall	Gorlin et al. 1990 Verloes, 1995	24	20%		PDA, BAV, MV abn	AVC	APVR	VSD, NS
Hydrolethalus	Salonen and Herva, 1990	>50	50%	TA		AVC		
Pseudotrisomy 13	Cohen and Gorlin, 1991 Verloes et al., 1991	25	60%	20% TOF, TGA	10% ASD, VSD, COA	25% AVC		40% VSD, NS
Silengo-Wachtel-Gardner	Greenberg et al., 1987	13	64%	>50% TOF, TGA, DORV				VSD, NS

* AoS, aortic stenosis; ASD, atrial septal defect; APVR, anomalous pulmonary venous return; AVC, atrioventricular canal; BAV, bicuspid aortic valve; COA, coarctation; DORV, double-outlet right ventricle; HLHS, hypoplastic left heart syndrome; MV, mitral valve; NS, not specified; PDA, patent ductus arteriosus; TA, truncus arteriosus; TGA, transposition of the great arteries; TOF, tetralogy of Fallot; VSD, ventricular septal defect.

evaluating a possible SLOS patient may be assisted by information about the pattern and frequency of CVMs in SLOS (Table VI). No CVM is pathgnomonic for SLOS (or any malformation syndrome), but atrioventricular canal-type CVMs, and anomalous pulmonary venous return, especially in combination, would support the diagnosis of SLOS.

Table VII reviews the small number of syndromes associated with an increase in class IV or V CVMs [Lin, 1990]. In none of these syndromes is there an increase in both class IV and V CVMs.

CONCLUSIONS

We conclude that CVMs are common (slightly less than 50%) in SLOS. Given the hemodynamic importance of most CVMs, all suspected patients should have a cardiac evaluation. Despite the overall predominance of "flow" CVMs, there is a striking increase in atrioventricular canal type defects and anomalous pulmonary venous return. Complex conotruncal CVMs are uncommon, and complex single-ventricle CVMs and heterotaxy have not yet been reported. The developmental error in SLOS, currently attributed to defective cholesterol biosynthesis, appears to favor alteration of the developmental mechanisms in atrioventricular canal (AVC) and anomalous pulmonary venous return (APVR).

Although CVMs in SLOS display mechanistic heterogeneity, in some instances the increased prevalence of AVC and APVR in SLOS, and the paucity of complex conotruncal or single-ventricle defects, may assist the dysmorphologist in differentiating SLOS from related syndromes.

TABLE VII. Syndromes Associated With Increased Prevalence of Class IV or V CVMs*

Syndrome	Frequency of CVM, all classes (%)	Frequency (%) of specific class	Specific CVMs
Down	40	60, class IV	Complete AVC ASD, primum type Canal-type/inlet VSD
Trisomy 18	99	20, class IV	Complete AVC Canal-type/inlet VSD
		99, class IV	Polyvalvar dysplasia
Ellis-van Creveld	50	75, class IV	Common atrium ASD primum Partial AVC
Cat-eye	40	40, class V	Total APVR
Ullrich-Turner	30	<5, class V	Partial APVR

* ASD, atrial septal defect; APVR, anomalous pulmonary venous return; AVC, atrioventricular canal; VSD, ventricular septal defect.

ACKNOWLEDGMENTS

We thank Magdalena Martin, Dr. Cynthia Curry, Dr. Sang Park, Michelle Fox, and Dr. Stephen Cedarbaum for their assistance.

REFERENCES

1. Abuelo DN, Tint GS, Kelley R, Batta AK, Shefer S, Salen G (1995): Prenatal detection of the cholesterol biosynthetic defect in the Smith-Lemli-Opitz syndrome by the analysis of amniotic fluid sterols. Am J Med Genet 56:281–285.
2. Akl KF, Khudr GS, Der Kaloustian VM, Najjar SS (1977): The Smith-Lemli-Opitz syndrome: Reports of a consanguineous Arab infant with bilateral focal renal dysplasia. Clin Pediatr (Phila) 16:665–668.
3. Ballesta F, Ferrer A, Serra M (1974): Q plus chromosome in 2 families with a Smith-Lemli-Opitz and Seckels syndrome. In Pearson PL, Lewis KR (eds): "Chromosomes Today," Vol 5. New York: Halsted/John Wiley.
4. Belmont JW, Hawkins E, Hetjmancik JF, Greenberg R (1987): Letter to the editor: Two cases of severe lethal Smith-Lemli-Opitz syndrome. Am J Med Genet 26:65–67.
5. Berry R, Wilson H, Robinson J, Sandlin C, Tyson W, Campbell J, Porreco R, Manchester D (1989): Apparent Smith-Lemli-Opitz syndrome and Miller-Dieker syndrome in a family with segregating translocation t(7;17)(q34;p13.1). Am J Med Genet 34: 358–375.
6. Bialer MG, Penchaszadeh VH, Kahn E, Libes R, Krigsman G, Lesser ML (1987): Female external genitalia and Müllerian duct derivatives in a 46,XY infant with the Smith-Lemli-Opitz syndrome. Am J Med Genet 28:723–731.
7. Blair HR, Martin JK (1966): A syndrome characterized by mental retardation, short stature, craniofacial dysplasia, and genital anomalies occurring in siblings. J Pediatr 69:457–459.
8. Buyse ML, Lai F (1990): Magnetic resonance imaging findings in an adult with Smith-Lemli-Opitz syndrome. Dysmorphol Clin Genet 4:145–148.
9. Casamassima AC, Mamunes P, Gladstone IM Jr, Solomon S, Moncure C (1987): A new syndrome with features of the Smith-Lemli-Opitz and Meckel-Gruber syndromes in a sibship with cerebellar defects. Am J Med Genet 26:321–336.
10. Cass D (1990): Aganglionosis: Associated anomalies. J Paediatr Child Health 26:351–354.
11. Chakanovskis JE, Sutherland GR (1971): The Smith-Lemli-Opitz syndrome in a profoundly retarded epileptic boy. J Ment Defic Res 15:153–162.
12. Chasalow FI, Blethen SL, Taysi K (1985): Possible abnormalities of steroid secretion in children with Smith-Lemli-Opitz syndrome and their parents. Steroids 46:827–843.
13. Cherstvoy ED, Lazjuk GI, Lurie IW, Nedzved MK, Usoev SS (1975): The pathological anatomy of the Smith-Lemli-Opitz syndrome. Clin Genet 7:382–387.
14. Cherstvoy ED, Lazjuk GI, Ostrovskaya TI, Shved IA, Kravtzova GI, Lurie IW, Gerasimoivh AI (1984): The Smith-Lemli-Opitz syndrome. A detailed pathological study as a clue to an etiological heterogeneity. Virchows Arch 404:413–425.
15. Chitayat D, Fagerstrom CL, Kalousek DK, Rootman J, Taylor GP, Hall JG (1989): De novo reciprocal 1p;2q translocation in a child with multiple congenital anomalies/mental retardation syndrome. Am J Med Genet 32:36–41.
16. Clark EB (1990): Growth, morphogenesis, and function. The dynamics of cardiac development. In Moller JH, Neal WA, (eds): "Fetal, Neonatal, and Infant Cardiac Disease." East Norwalk: Appleton and Lange, pp 3–23.
17. Cohen MM, Gorlin RJ (1991): Pseudo-trisomy 13 syndrome. Am J Med Genet 39:332–335.
18. Cotlier E, Rice P (1971): Cataracts in the Smith-Lemli-Opitz syndrome. Am J Ophthalmol 72:955–959.
19. Cunniff C, Kratz LE, Moser A, Natowicz MR, Kelley RI (1997): Clinical and biochemical spectrum of patients with RSH/Smith-Lemli-Opitz syndrome and abnormal cholesterol metabolism. Am J Med Genet 68:270–278.
20. Curry CJR, Carey JC, Holland JS, Chopra D, Fineman R, Golabi M, Sherman S, Pagon RA, Allanson J, Shulman S, Barr M, McGravey V, Dabiri C, Schimke N, Ives E, Hall BD (1987): Smith-Lemli-Opitz syndrome type II: Multiple congenital anomalies with male pseudohermaphroditism and frequent early lethality. Am J Med Genet 26:45–57.
21. Dallaire L, Fraser FC (1966): The syndrome of retardation with urogenital and skeletal anomalies in siblings. J Pediatr 69: 459–460.
22. Dallaire L, Fraser FC (1969): The Smith-Lemli-Opitz syndrome of retardation, urogenital and skeletal anomalies. In Bergsma D (ed): "The First Conference on The Clinical Delineation of Birth Defects. Part II: Malformation Syndromes." New York: The National Foundation—March of Dimes. BD:OAS V (2):180–182.
23. Donnai D, Young ID, Owen WG, Clark SA, Miller PFW, Knox WF (1986): The lethal multiple congenital anomaly syndrome of polydactyly, sex reversal, renal hypoplasia, and unilobar lungs. J Med Genet 23:64–71.
24. Donnai D, Burn J, Hughes H (1987): Letter to the editor: Smith-Lemli-Opitz syndromes: Do they include the Pallister-Hall syndrome? Am J Med Genet 28:741–743.
25. Donnenfeld AE, Zackai EH, McDonald DM, Aquino R, Emanuel BS (1987): De novo 2q+ masquerading as Smith-Lemli-Opitz syndrome. J Med Genet 24:436–439.
26. Ferencz C, Rubin JD, McCarter RJ, Boughman JA, Wilson PD, Brenner JI, Neill CA, Perry LW, Hepner SI, Downing JW (1987): Cardiac and noncardiac malformations: Observations in a population-based study. Teratology 35:367–378.
27. Fine RN, Gwinn JL, Young EF (1968): Smith-Lemli-Opitz syndrome. Radiologic and postmortem findings. Am J Dis Child 115: 483–488.
28. Finley SC, Finley WH, Monsky DB (1969): Cataracts in a girl with features of the Smith-Lemli-Opitz syndrome. J Pediatr 75: 706–707.
29. Freedman RA, Baum JL (1979): Postlenticular membrane associated with Smith-Lemli-Opitz syndrome. Am J Ophthalmol 87: 675–677.
30. Fried K, Fraser WI (1972): Smith-Lemli-Opitz syndrome in an adult. J Ment Defic Res 15:30–34.
31. Fukazawa R, Nakahori Y, Kogo T, Kawakami T, Akamatsu H, Tanae A, Hibi I, Nagafuchi S, Nakagome Y, Hirayama T (1992): Normal Y sequences in Smith-Lemli-Opitz syndrome with total failure of masculinization. Acta Paediatr 81:570–572.
32. Garcia CA, McGarry PA, Voirol M, Duncan C (1973): Neurological involvement in the Smith-Lemli-Opitz syndrome: Clinical and neuropathological findings. Dev Med Child Neurol 15:46–55.
33. Gibson R (1965): A case of the Smith-Lemli-Opitz syndrome of multiple congenital anomalies in association with dysplasia epiphysialis punctata. Can Med Assoc J 92:574–575.
34. Gold JD, Pfaffenbach DD (1975): Ocular abnormalities in the Smith-Lemli-Opitz syndrome. J Pediatr Ophthalmol 12:228–234.
35. Gorlin RJ, Cohen MM Jr, Levin LS (1990): "Syndromes of the Head and Neck." New York: Oxford University Press pp684–685, 903–905.
36. Greenberg F, Gresik MV, Carpenter RJ, Law SW, Hoffman LP, Ledbetter DH (1987): The Gardner-Silengo-Wachtel or genitopalato-cardiac syndrome: Male pseudohermaphroditism with micrognathia, cleft palate, and conotruncal cardiac defect. Am J Med Genet 26:59–64.
37. Greene C, Pitts W, Rosenfeld R, Luzzatti L (1984): Smith-Lemli-Opitz syndrome in two 46,XY infants with female external genitalia. Clin Genet 25:366–372.
38. Hanissian AS, Summitt RL (1969): Smith-Lemli-Opitz syndrome in a Negro child. J Pediatr 74:303–305.
39. Harbin RL, Katz JI, Frias JL, Rabinowicz IM, Kaufman HE (1977): Sclerocornea associated with the Smith-Lemli-Opitz syndrome: Am J Ophthalmol 84:72–74.
40. Herman TE, Siegel MJ, Lee BCP, Dowton SB (1993): Smith-Lemli-Opitz syndrome type II: Report of a case with additional radiographic findings. Pediatr Radiol 23:37–40.
41. Hill S, Creasy M, Bundey S (1991): A family with 3 sisters with the 4p− syndrome, originally reported as suffering from the Smith-Lemli-Opitz syndrome. J Ment Defic Res 35:76–81.

42. Hobbins JC, Jones OW, Gottesfeld S, Persutte W (1994): Transvaginal ultrasonography and transabdominal embryoscopy in the first-trimester diagnosis of Smith-Lemli-Opitz syndrome, type II. Am J Obstet Gynecol 171:546–549.
43. Hoefnagel D, Wurster D, Pomeroy J, Benz R (1969): The Smith-Lemli-Opitz syndrome in an adult male. J Ment Defic Res 13: 249–257.
44. Hummel M, Baker JC, Graham SF (1991): Further delineation of a severe form of Smith-Lemli-Opitz syndrome. Pediatr Res 29:69.
45. Irons M, Elias ER, Tint GS, Salen G, Frieden R, Buie TM, Ampola M (1994): Abnormal cholesterol metabolism in the Smith-Lemli-Opitz syndrome: Report of clinical and biochemical findings in four patients and treatment in one patient. Am J Med Genet 50:347–352.
46. Johnson VP (1975): Smith-Lemli-Opitz syndrome: Review and report of two affected siblings. Z Kinderheilkd 119:221–234.
47. Johnson JA, Aughton DJ, Comstock CH, von Oeyen PT, Higgins JV, Schulz R (1994): Prenatal diagnosis of Smith-Lemli-Opitz syndrome, type II. Am J Med Genet 49:240–243.
48. Joseph DB, Uehling DT, Gilbert E, Laxova R (1987): Genitourinary abnormalities associated with the Smith-Lemli-Opitz syndrome. J Urol 137:719–721.
49. Judge CG, Chakanovskis JE, Sutherland GR (1971): The Smith-Lemli-Opitz syndrome. Med J Aust 2:145–147.
50. Kelley RI (1995): Diagnosis of Smith-Lemli-Opitz syndrome by gas chromatography/mass spectrometry of 7-dehydrocholesterol in plasma and amniotic fluid and cultured skin fibroblasts. Clin Chem Acta 236:45–58.
51. Kenis H, Hustinx TWJ (1971): A familial syndrome of mental retardation in association with multiple congenital anomalies resembling the syndrome of Smith-Lemli-Opitz. Maandschr Kindergeeneeskd 35:37–48.
52. Kim EH, Boutwell WC (1985): Letter: Smith-Lemli-Opitz syndrome associated with Hirschsprung disease, 46,XY female karyotype and total anomalous pulmonary venous drainage. J Pediatr 106:861.
53. Kohler HG (1983): Brief clinical report: Familial neonatally lethal syndrome of hypoplastic left heart, absent pulmonary lobation, polydactyly, and talipes, probably Smith-Lemli-Opitz (RSH) syndrome. Am J Med Genet 14:423–428.
54. Kretzer FL, Hittner HM, Mehta RS (1981): Ocular manifestations of the Smith-Lemli-Opitz syndrome. Arch Ophthalmol 99:2000–2006.
55. Lachman MF, Wright Y, Whiteman DA, Herson V, Greenstein RM (1991): Brief clinical report. A 46,XY phenotypic female with Smith-Lemli-Opitz syndrome. Clin Genet 39:136–141.
56. Lacombe D, Beon C, Lobaccaro JM, Sultan C, Battin J (1991): Androgen receptor study in one case of the Smith-Lemli-Opitz syndrome. Proceedings of the Eighth International Congress of Human Genetics. Am J Hum Genet [Suppl] 49:146.
57. LeMerrer M, Briard ML, Girard S, Mulliez N, Moraine C, Imbert MC (1988): Lethal acrodysgenital dwarfism: A lethal condition resembling Smith-Lemli-Opitz syndrome. J Med Genet 25: 88–85.
58. Lin AE (1990): Congenital heart defects in malformation syndromes. Perinatol Clin N Am 17:641–673.
59. Lipson A, Hayes A (1984): Letter: Smith-Lemli-Opitz syndrome and Hirschspurng disease. J Pediatr 105:177.
60. Lowry RB (1983): Editorial comment: Variability in the Smith-Lemli-Opitz syndrome: Overlap with the Meckel syndrome. Am J Med Genet 14:429–433.
61. Lowry RB, Yong S-L (1980): Borderline normal intelligence in the Smith-Lemli-Opitz (RSH) syndrome. Am J Med Genet 5: 137–143.
61. Lowry RB, Miller JR, MacLean JR (1968): Micrognathia, polydactyly, and cleft palate. J Pediatr 72:859–861.
63. Marion RW, Alvarez LA, Marans ZS, Lantos G, Chitayat D (1987): Computed tomography of the brain in the Smith-Lemli-Opitz syndrome. J Child Neurol 2:198–200.
64. McGaughran JM, Clayton PT, Mills KA, Rimmer S, Moore L, Donnai D (1995): Prenatal diagnosis of Smith-Lemli-Opitz syndrome. Am J Med Genet 56:269–272.
65. McKeever PA, Young ID (1990): Smith-Lemli-Opitz syndrome II: A disorder of the fetal adrenals? J Med Genet 27:465–466.
66. Meinecke P, Blunck W, Rodewald A (1987): Letter to the editor: Smith-Lemli-Opitz syndrome. Am J Med Genet 28:735–739.
67. Nakai H, Satoh T, Otomo H, Igarashi Y, Omura K, Tada K (1980): A case of Smith-Lemli-Opitz syndrome with first cousin parents. Teratology 22:39A.
68. Nevo S, Benderly A, Levy J, Katznelson MBM (1972): Smith-Lemli-Opitz syndrome in an inbred family. Am J Dis Child 124:431–433.
69. Nwokoro N, Hyde B, Mulvihill JJ (1994): Smith-Lemli-Opitz syndrome: Biochemical before clinical diagnosis; early dietary management. Am J Med Genet 50:375–376.
70. Opitz JM, Howe JJ (1969): The Meckel-Gruber syndrome (dysencephalia splanchnocystica, the Gruber syndrome). In Bergsma D (ed): "The First Conference on the Clinical Delineation of Birth Defects. Part II: Malformation Syndromes." New York: The National Foundation—March of Dimes. BD:OAS V (2):167–179.
71. Opitz JM, Zellweger H, Shannon WR, Ptacek LJ (1969): The RSH syndrome. In Bergsma D (ed): "The First Conference on the Clinical Delineation of Birth Defects. Part II: Malformation Syndromes." New York: The National Foundation—March of Dimes. BD:OAS V (2):43–52.
72. Opitz JM, Penchaszadeh VB, Holt MC, Spano LM, Smith VL (1994): Smith-Lemli-Opitz (RSH) syndrome bibliography: 1964–1993. Am J Med Genet 50:339–343.
73. Pankau R, Partsch CJ, Funds J, Sippell WG (1992): Hypothalamic-pituitary-gonadal function in two infants with Smith-Lemli-Opitz syndrome. Am J Med Genet 43:513–516.
74. Park SC, Needles CF, Dimich I, Sussman L (1968): Congenital heart disease in an infant with the Smith-Lemli-Opitz syndrome. J Pediatr 73:896–902.
75. Parnes S, Hunter AG, Jimenez C, Carpenter BF, McDonald I (1990): Apparent Smith-Lemli-Opitz syndrome in a child with a previously undescribed form of mucolipidosis not involving the neurons. Am J Med Genet 35:397–405.
76. Patterson K, Toomy KE, Chandra RS (1983): Hirschsprung disease in a 46,XY phenotypic infant girl with Smith-Lemli-Opitz syndrome. J Pediatr 103:425–427.
77. Pierquin G, Peeters P, Roels F, Vamos E, Brucher JM, Tint GS, Honda A, Van Regemorter N (1995): Severe Smith-Lemli-Opitz syndrome with prolonged survival and lipid abnormalities. Am J Med Genet 56:276–280.
78. Pinsky L, DiGeorge AM (1965): A familial syndrome of facial and skeletal anomalies associated with genital abnormality in the male and normal genitals in the female. J Pediatr 66:1049–1054.
79. Robinson D, Perry LW, Barlee A, Mella GW (1971): Smith-Lemli-Opitz syndrome with cardiovascular abnormality. Pediatrics 47: 844–847.
80. Rossiter JP, Hofman KJ, Kelley RI (1995): Smith-Lemli-Opitz syndrome: Prenatal diagnosis by quantification of cholesterol precursors in amniotic fluid. Am J Med Genet 56:272–275.
81. Rudolph C, Benaroch L (1995): Hirschsprung disease. Pediatr Rev 16:5–11.
82. Rutledge JC, Friedman JM, Harrod MJE, Currarino G, Wright CG, Pinckney L, Chen H (1984): A "new" lethal multiple congenital anomaly syndrome. Joint contractures, cerebellar hypoplasia, renal hypoplasia, urogenital anomalies, tongue cysts, shortness of limbs, eye abnormalities, defects of the heart, gallbladder agenesis, and ear malformations. Am J Med Genet 19:255–264.
83. Ruvalcalba RHA, Reichert A, Smith DW (1968): Smith-Lemli-Opitz syndrome. Case report. Arch Dis Child 43:620–623.
84. Salonen R (1984): The Meckel syndrome: Clinicopatholgical findings in 67 patients. Am J Med Genet 18:671–689.
85. Salonen R, Herva R (1990): Hydrolethalus syndrome. J Med Genet 27:756–759.
86. Saraiva JM, Baraitser M (1992): Joubert syndrome: A review. Am J Med Genet 43:726–731.
87. Scarbrough PR, Huddleston K, Finley SC (1986): An additional case of Smith-Lemli-Opitz syndrome in a 46,XY infant with female external genitalia. J Med Genet 23:174–175.
88. Seller MJ, Russell J, Tint GS (1995): Unusual case of Smith-Lemli-Opitz syndrome "type II." Am J Med Genet 56:265–268.

89. Shen-Schwartz S, Dave H (1988): Meckel syndrome with polysplenia: Case report and review of the literature. Am J Med Genet 31:349–355.

90. Sinclair L, Winterborn MH, Brown J (1969): Smith-Lemli-Opitz syndrome. Proc R Soc Med 62:907–908.

91. Singer LP, Marion RW, Li JKH (1989): Limb deficiency in an infant with Smith-Lemli-Opitz syndrome. Am J Med Genet 32: 380–383.

92. Smith DW, Lemli L, Opitz JM (1964): A newly recognized syndrome of multiple congenital anomalies. J Pediatr 64:210–217.

93. Suzumori K, Koike K, Yagami Y (1974): Diagnosis of fetal abnormality by fetography. Teratology 10:98–99.

94. Thompson E, Baraitser M (1987): Letter to the editor: Nosology of Smith-Lemli-Opitz syndrome. Am J Med Genet 28:733–734.

95. Tint GS, Irons M, Elias ER, Batta AK, Frieden R, Chen TS, Salen G (1994): Defective cholesterol biosynthesis associated with the Smith-Lemli-Opitz syndrome. N Engl J Med 330:107–113.

96. Tzouvelekis G, Antoniades K, Batma A, Nanas C (1991): Smith Lemli-Opitz syndrome in female, monozygotic twins. Clin Genet 40:229–232.

97. Verloes A (1995): Numerical syndromology: A mathematical approach to the nosology of complex phenotypes. Am J Med Genet 55:433–443.

98. Verloes A, Aymé S, Gambarelli D, Gonzales M, Le Merrer M, Mulliez N, Philip N, Roume J (1991): Holoprosencephaly-polydactyly ("pseudotrisomy 13") syndrome: A syndrome with features of hydrolethalus and Smith-Lemli-Opitz syndromes. A collaborative multicentre study. J Med Genet 28:297–303.

99. Wallace M, Zori RT, Alley T, Whidden E, Gray BA, Williams CA (1994): Smith-Lemli-Opitz syndrome in a female with a de novo, balanced translocation involving 7q32: Probable disruption of an SLOS gene. Am J Med Genet 50:368–374.

100. Yunis E, Gonzalez J, Zuniga R, Torres de Caballero OM, Mandragon A (1979): Clinical case reports: Direct duplication 2p14–2p23. Hum Genet 47:241–244.

101. Zahle Ostergaard G, Nielsen H, Friis N (1992): Defective monocyte oxidative metabolism in a child with Smith-Lemli-Opitz syndrome. Eur J Pediatr 151:291–294.

102. Zizka J, Maresová J, Kerekes Z, Nozicka Z, Juttnerová V, Baliček P (1983): Case report: Intestinal aganglionosis in the Smith-Lemli-Opitz syndrome. Acta Paediatr Scand 72:141–143.